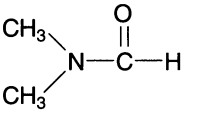
N,N-Dimethylformamide

CAS #68-12-2
Swiss CD-1 mice, at 0.0, 1000, 4000, and 7000 ppm in drinking water
Jerrold Heindel, NTP/NIEHS Project Officer
Julia George, Patricia Fail, Tom Grizzle, and Donald Feldman
Research Triangle Institute
Started 9/11/90; Completed 9/24/92
NTIS #PB92123842



N,N-Dimethylformamide (DMF) was tested as part of a structure–activity study, using the RACB protocol and Swiss CD-1 mice, to generate some public-domain reproduction data on this high volume chemical with known developmental toxicity. Data on food and water consumption, body weights, and clinical signs during a 2-week dose-range-finding study (Task 1) were used to set exposure concentrations for the Task 2 continuous cohabitation study at 1000, 4000, and 7000 ppm in drinking water. Based on mean body weight and average water consumption data, these concentrations produced estimated daily consumptions of approximately 200, 700, and 1200 mg/kg/day, respectively.

In the F_0 animals, there was a progressive decrease in fertility that was faster in the high dose and middle dose animals than was observed in the controls (90 → 55%, 100 → 70%, and 100 → 92%, respectively). The average number of litters per pair was reduced in the middle and high dose groups by 8 and 22%, compared to controls, while the average number of pups per litter was reduced by 36 and 55%, respectively. Concomitantly, the average live pup weight adjusted for litter size was reduced in these two dose groups by 18 and 21%.

Pups born to DMF-treated pairs had deformations, which included domed heads and hematomas on the head and face. The proportions of litters with greater than or equal to 1 abnormal pup (from control to high dose) were 8, 11, 90, and 80%, respectively. The slight decrease at the top dose group is attributed to cannibalism of the most-affected pups.

The cumulative days to deliver each litter was increased only at the high dose and only statistically for litter 3, though all other means at this dose level were greater than control. While postpartum sire weights were never reduced by DMF, dam weights after delivery of the 5th litter were reduced in all treated groups by 7, 8, and 9%, from low to high dose. Female feed consumption was increased by 10 to 16% in the middle and high-dose groups.

Pups from the last litter were reared by the dam until weaning, and then consumed the same levels of DMF provided to their parents. There were dose-related reductions in feed and water consumption by the dams during nursing, from 13 to approximately 30%. There was increased postnatal mortality in pups in the middle and high dose groups but no reductions in pup weight gain pre-weaning.

After weaning this last litter, a crossover mating trial was conducted using the control and high dose mice. No statistical differences from control were found, although the litters delivered to 7000 ppm DMF-treated females were 32% smaller than those delivered to control pairs. Skeletal and soft-tissue examination of pups born during Task 3 found that pups born to DMF-treated dams showed the same spectrum of malformations as seen in Task 2, and these litters also had more skeletally malformed fetuses per litter than controls (95 vs 40%).

After the Task 3 litters were delivered, evaluated, and killed, the F_0 mice were killed and necropsied. There was no significant difference between male body weights, though the high dose group weighed approximately 8% less than controls. Relative liver weights in males were increased by 24, 52, and 52% in the low, middle, and high dose groups, respectively. These same groups also had increased caudal epididymis weights (31, 23, and 26%, respectively), while only the high dose group showed an increase in relative kidney weights (15%). There were no changes in sperm end points, although testicular spermatid head count was reduced in the low and high dose groups by 25 and 19%, respectively. For F_0 females, high dose body weights were reduced by 11%, while in the low, middle, and high dose groups, relative liver weight was increased by 31, 46, and 35%, respectively, while relative kidney weight was increased by 7, 7, and 13%. There was no difference between the control and high dose females in estrous parameters. There were no significant dose-related microscopic findings in the F_0 mice.

All dose groups and controls were evaluated in the F_1 mating trial where only half of the cohabited pairs in the middle and high dose groups had a litter, versus 90% of controls. Dam weights were reduced by 14 and 18% in the middle and high dose groups, respectively. Also in these two groups, litter size was reduced by 57 and 64%, respectively, while adjusted pup weight was reduced in the low to high groups, by 6, 25, and 24%, respectively. In the control, low, middle, and high dose groups, the proportion of pups that showed similar malformations to those seen earlier were 0, 28, 60, and 75%.

After the F_2 pups were delivered, evaluated, and killed, the F_1 adults were killed and necropsied. In males, there was a 10 and 6% reductions in body weight seen in the middle and high dose groups, respectively. From the low to high dose groups, liver weight was increased by 37, 54, and 57%, and prostate weight was reduced by 13, 15, and 24%, respectively. There was a monotonic decrease in epididymal sperm concentration which reached statistical significance at the high dose (20% reduced); no other sperm measure was affected.

In females, body weight in the middle and high dose groups was reduced by 12 and 12%, while relative kidney weights were increased by 10% in both groups. Relative liver weights were increased in

dams from the low to high dose groups by 23, 37, and 34%, respectively. Estrous cycle length was significantly increased in high dose treated females, and fewer of these females had cycles. In both sexes, microscopic findings were minimal and not strongly related to dose.

These data show that N,N-dimethylformamide at greater than or equal to 4000 ppm reduced fertility, and at greater than or equal to 1000 ppm increased malformations in offspring. These effects occurred concomitant with increases in liver weights, but at doses lower than those required to reduce body weight. Although the female appears to be the most affected sex, data from the crossover mating trial also are consistent with these effects being couple-dependent, as there was no clear fertility difference between the sexes.

Summary: NTP Reproductive Assessment by Continuous Breeding Study.

NTIS#: **PB92123842**
Chemical: **N,N-Dimethylformamide**
CAS#: **68-12-2**
Mode of exposure: **Drinking water**
Species/strain: **Swiss CD-1 mice**

F_0 generation — Dose concentration →	1000 ppm	4000 ppm	7000 ppm
General toxicity	Male, female	Male, female	Male, female
Body weight	—, —	—, —	—, ↓
Kidney weight[a]	—, ↑	—, ↑	↑, ↑
Liver weight[a]	↑, ↑	↑, ↑	↑, ↑
Mortality	—, —	—, —	—, —
Feed consumption	—, —	—, ↑	—, ↑
Water consumption	—, —	—, —	—, —
Clinical signs	—, —	↑, ↑	↑, ↑
Reproductive toxicity			
x̄ litters/pair	—	↓	↓
# live pups/litter; pup wt./litter	—, —	↓, ↓	↓, ↓
Cumulative days to litter	—	—	↑
Absolute testis, epididymis weight[a]	—, ↑	—, ↑	—, ↑
Sex accessory gland weight[a] (prostate, seminal vesicle)	—, —	—, —	—, —
Epidid. sperm parameters (#, motility, morphology)	—, —, —	—, —, —	—, —, —
Estrous cycle length	•	•	—

Determination of affected sex (crossover)	Male	Female	Both
Dose level	—	—	—

F_1 generation — Dose concentration →	1000 ppm	4000 ppm	7000 ppm
General toxicity	Male, female	Male, female	Male, female
Pup growth to weaning	—, —	—, —	—, —
Mortality	—, —	↑, ↑	↑, ↑
Adult body weight	—, —	↓, ↓	↓, ↓
Kidney weight[a]	—, —	—, ↑	—, ↑
Liver weight[a]	↑, ↑	↑, ↑	↑, ↑
Feed consumption	—, —	—, —	—, —
Water consumption	—, —	—, —	↑, —
Clinical signs	—, —	↑, ↑	↑, ↑
Reproductive toxicity			
Fertility index	—	—	—
# live pups/litter; pup wt./litter	—, ↓	↓, ↓	↓, ↓
Absolute testis, epididymis weight[a]	—, —	—, —	—, —
Sex accessory gland weight[a] (prostate, seminal vesicle)	↓, —	↓, —	↓, —
Epidid. sperm parameters (#, motility, morphology)	—, —, —	—, —, —	↓, —, —
Estrous cycle length	•	•	↑

Summary information	
Affected sex?	Unclear
Study confounders:	None
NOAEL reproductive toxicity:	<1000 ppm
NOAEL general toxicity:	<1000 ppm
F_1 more sensitive than F_0?	No
Postnatal toxicity:	Yes

Legend: —, no change; •, no observation; ↑ or ↓, statistically significant change ($p<0.05$); —, —, no change in males or females. [a]Adjusted for body weight.